## OUTPUT 5
## Activities related with the maintenance of the germplasm bank of cassava and other *Manihot* species.

CIAT has been trusted with the maintenance of the cassava world germplasm bank, which includes more than 6000 accessions of *Manihot esculenta* and other *Manihot* species. In the following pages a summary of activities related to the germplasm bank will be described.

### *Activity 5.1. Maintenance of Manihot germplasm bank in the field.*

**Rationale**

The Genetic Resources Unit is officially in charge of the maintenance of the cassava germplasm bank, both *in vitro* and in the field. However, for practical reasons, the field operations are coordinated by IP3 project. Since year 2000 an extensive activity to clean up from frogskin disease, the germplasm bank has been carried out. Plots from the germplasm bank maintained in the field, because of its very nature, could not be eliminated even if frogskin disease appeared in some of the plants. Eventually the incidence of the disease increased to unacceptable levels.

**Specific Objectives**:

a) *To contribute with the maintenance the cassava and related wild species germplasm bank in the field.*
b) *Implementation of scheme for reducing incidence of frogskin disease in germplasm bank and elite clones.*

**Materials and methods**

Drastic measures were taken to reduce the level of incidence of frogskin disease back to acceptable levels. The strategy implies four main elements:

<u>Regeneration of each accession from the in vitro collection</u>. From each accession, a plant from the *in vitro* collection was regenerated and indexed to certify it is free of diseases. Plants passing this first test are then hardened in conditions that do not allow for the presence of white flies, and therefore, minimizes the possibility of acquiring the frogskin disease agent again.

<u>*Planting of disease free plantlets outside CIAT, in isolated fields*</u>. Because of the higher incidence of frogskin disease at CIAT (mainly at the germplasm bank collection in the field), plants that are certified to be disease free, or those developed from botanical seeds (which do not transmit viral agents to the plants germinating from them), were planted outside CIAT in isolated plots (CEUNP). Only virus-free plants were planted in those isolated plots. In the meantime, plantings at CIAT were reduced as a higher proportion of the cassava germplasm is being certified to be disease-free. In short the outside plantings were certified to be *"clean",* whereas the plantings at CIAT were not. This situation was maintained until the middle of

2001, when materials not certified to be disease free moved out of CIAT, and those that are *clean*, came back to the station.

*Breaking the life cycle of the white flies at CIAT*. In addition of maintaining an ideal reservoir for the agent of the frogskin disease in the germplasm bank, there is a second factor that facilitated the spread of the disease. In effect, the white flies problem has increased considerably during the last few years. A major factor for this increment has been the continuous planting of cassava year round. The insects, therefore, had an ideal condition for maintaining high population densities. Between July 8 and August 9, 2001, there was no cassava plant in the field at CIAT's station in Palmira. It is expected that this measure will reduce population densities for the insect, and in turn, will reduce to a minimum the already inefficient transmission of the frogskin disease agent to healthy plants.

*Harvest of stakes only from asymptomatic plants*. A common procedure to harvest cassava is to first take the stakes (vegetative *"seed"*) out of the field, and then harvest the roots. In fact this practice prevents the elimination of stakes from diseased plants, because when the roots are evaluated for symptoms, the stakes from each plant has already been mixed with other stakes from different plants. Starting in this year, the harvest protocol has been changed slightly. The whole plant is first taken out of the ground, so before taking the stakes the roots can be inspected to make sure they are asymptomatic. Stakes are taken only from plants that do not show the symptoms. This practice will reduce to a very minimum the "seed" transmission of the disease to only two possible cases: **a)** when the worker fails to recognize the symptoms; or **b)** when the plant has been infected late in the season and, therefore, it does not show the symptoms but the disease will be transmitted through its stakes.

**Results**

All the activities were carried out as expected. A large proportion of accessions from the germplasm bank was evaluated for frogskin disease and, if clean, planted in isolated conditions. Sequential plantings were performed as the plants were certified to be disease-free. Therefore, harvest of these plants was also done sequentially. The levels of frogskin were very low, as expected. However, given the results from the previous year, when higher than acceptable levels of frogskin disease were observed, it has been decided not to plant the entire germplasm bank in the field, until the vector(s) and pathogen(s) are clearly determined.

During the current year the possibility of a viroid related to frogskin disease was discarded. Currently a viral agent and/or a phytoplasm have been postulated as the causal agent of this elusive disease (See Output 7). Scientists working in cassava entomology are also carrying out studies to determine which species may be acting as a vector of the disease. Although it has always been suspected that whiteflies may be involved, now the possibility of a cycadelid is also considered.

A major achievement was in relation to the management of the whiteflies problem. By interrupting the breeding cycle of the insect by eliminating all cassava plants in the field for one month, we have achieved excellent results. By November, cassava plantings in the station were still remarkably clean from whiteflies and only at the end of the season we could see some population growth. As a result, we have seen, for the first time in many years, other arthropods pest coming back. That was the case of mites and mealybugs. This is, in a way, an evidence of the success we have had reducing the problem of the whiteflies that reached such a high level of incidence that prevented other pest colonizing cassava plantings at CIAT-Palmira.

### *Activity 5.2. Evaluation of M. esculenta and related species from the germplasm collection for useful traits, particularly for the natural occurrence of apomixis.*

**Rationale**

Apomixis is a highly desirable trait for cassava. This mode of reproduction would facilitate germplasm exchange because the shipment of botanical seed implies much lower phytosanitary risks than vegetative propagules. Apomictic seed could also be used to conserve germplasm for longer period of time a much lower costs. Also, depending on the genoytpe, apomixis could greatly increase multiplication rate once the "seed" of an elite clone needs to be increased.

**Specific Objectives**:

a) *To search for the natural occurrence of apomixis in the germplasm bank collection.*
b) *To carry out collaborative research with other institutions in the area of apomixis.*

**Materials and methods**

Apomixis has always been an interesting process to cassava breeders. There have been some reports of apomixis occurrence in the *Manihot* genus (Nassar et al., 1998. Genetics and Molecular Biology 21:527-530). This reproductive abnormality is likely to occur in cassava germplasm introgressed with wild species. The germplasm bank includes accessions collected in areas where the likelihood of natural crossing between *M. esculenta* and other *Manihot* species is high (especially from the Amazon basin). Therefore, we have already started to bag clusters of female flowers, searching for a genotype that will produce seed without pollination. Having apomictic cassava would greatly simplify maintaining genetic stocks unchanged, and also facilitate the exchange of germplasm almost without the risks of introducing diseases.

**Results**

So far the process has yielded no positive results, but only a fraction of the entire germplasm bank collection has been screened, because of the transitional stage most of the accessions were going through to be cleaned from frogskin disease. This activity, which has low probability of success, will be maintained until the entire collection has been properly tested.

### *Activity 5.3. Evaluation of M. esculenta and related species from the germplasm collection for novel types of starches and other traits of economic relevance.*

**Rationale**

The starch industry has always requested for different types of starches in cassava roots. No extensive evaluation has been made on starch properties of each accession in the germplasm bank. Novel starch types have large economic relevance for the industry.

Protein levels in cassava roots are low. This situation results in the price of flour or chips from dried cassava roots being about 70% of that of field maize. Increasing protein levels in cassava roots is therefore an important objective for the industrial processing of cassava for animal feed, and certainly a critical goal for those areas where cassava is important as a food security crop.

**Specific Objectives**:

a) *To screen the germplasm bank in search for the natural occurrence of novel starch types*
b) *To search for high-protein cassava roots.*

**Results**

We have begun a systematic characterization of the starch properties in the roots of the accessions from the germplasm bank. Every year up to 2000 accessions are evaluated.

Also based on the results from a group of 600 genotypes we have determined the possibility of higher protein levels in cassava roots originated from Meso-America, particularly Guatemala. We are recovering clones from that region of the world to confirm this hypothesis.